論文内容の要旨

Synergistic Enhancement of Cancer Therapy Using Combination of Hyperthermia with Physical and Chemical Modifiers

MONIRUZZAMAN
Department of Oral and Maxillofacial Surgery
Graduate School of Medicine & Pharmaceutical Sciences
University of Toyama, Japan
March, 2019

Thesis Abstract

Cancer is still the leading cause of human morbidity and mortality worldwide, with increasing incidence because of changing lifestyle and increased exposure to carcinogens. Most of the available treatments like surgery, chemotherapy, and radiotherapy are associated with undesirable side effects. Thus, explore for more selective anti-cancer strategy should be urgently required. Hyperthermia (HT) is a promising therapeutic tool which acts by directly damaging and killing cancer cells or enhancing the efficacy of other existing cancer treatment modalities (e.g. radiotherapy, chemotherapy, etc.) against various cancer types, with minimal injury to normal cells. However, the efficiency of hyperthermia treatment is related to the temperature achieved during the heating, the exposure time, and tumor cell characteristics. Therefore, in many circumstances, its cytotoxic effect is often insufficient for quantitative cancer cell

death because of these biological and technical problems. To overcome these challenges, several studies have explored non-toxic enhancers for HT-induced cell death. Here we employed HT as an adjuvant therapy with cold atmospheric helium plasma (He-CAP), which produces an enormous amount of reactive oxygen species (ROS) in the liquid phase, and an anti-inflammatory drug, 5-aminosalicylic acid (5-ASA) that clinically use in the treatment of inflammatory bowel disease (IBD). In this study, we addressed how HT enhances He-CAP and 5-ASA induced cell death in various cancer cells. Combined treatment of HT with He-CAP and 5-ASA significantly enhanced apoptosis and cytotoxicity in cancer cells, but not in normal cells. Furthermore, the combination was associated with increases of reactive oxygen and nitrogen species (ROS/RNS) generation, endoplasmic reticulum (ER) stress marker proteins expression, intracellular calcium $[Ca^{2+}]i$ concentrations. Apoptotic endpoints were significantly increased by the combination treatment, as evidenced by the presence of Annexin-V/PI-positive cells, loss of mitochondrial membrane potential, Bcl-2/Bax ratio alteration, increase in the expression levels of the death receptor Fas and cleaved Bid, and caspase activation. Interestingly, the enhancement of apoptosis was reversed in the presence of ROS/RNS scavengers. These findings indicate that the HT in combination with He-CAP and 5-ASA synergistically enhances apoptosis via ROS/RNS-mediated ER stress-Ca^{2+}-mitochondria signaling and caspase-dependent apoptotic pathways. Our findings provide novel evidence that HT could be an enhancing agent for the physical modality (He-CAP) and chemical modality (5-ASA) in various tumors. More importantly, it reveals that increased ROS/RNS generation might be an effective strategy in treating human cancer. In conclusion, this work provides evidence for the novel anti-cancer strategy which might be used for the treatment of cancer in the future and further in vivo studies are needed.

Keywords: Hyperthermia, Cold atmospheric helium plasma, 5-Aminosalicylic acid, Reactive oxygen species, Reactive nitrogen species, Cancer, and Apoptosis.